

WHAT IS CLAIMED IS:

1. ~~In a method for the production of a (protease) differing in at least one amino acid from a wild-type protease employing as an expression host a Bacillus strain incapable of producing (a) wild-type protease, the improvement which comprises:~~

using as an expression host a Bacillus strain wherein a first DNA sequence capable of homologous recombination with a second DNA sequence encoding said protease has been deleted from the chromosome of said expression host prior to transformation of a parent of said expression host with at least one copy of an integration vector comprising said second DNA sequence.

2. ~~The method according to Claim 1, wherein said Bacillus strain is an alkalophilic Bacillus strain.~~

3. ~~The method according to Claim 1, wherein said first DNA sequence is a wild-type protease gene.~~

4. ~~The method according to Claim 1, wherein said Bacillus strain is Bacillus novo species PB92.~~

5. ~~The method according to Claim 1, wherein said Bacillus strain is an asporogenic alkalophilic Bacillus strain.~~

6. ~~The according to Claim 5, wherein said first DNA sequence has been deleted (by) homologous or illegitimate recombination.~~

7. ~~The method according to Claim 2, wherein a plasmid comprises said second DNA sequence.~~

8. ~~The method according to Claim 7, wherein said protease is a high alkaline protease.~~

9. ~~The method according to Claim 8, wherein said protease is a mutation of a wild-type high alkaline protease having an amino acid sequence at least substantially similar to that of a Bacillus novo species PB<sub>92</sub> protease or a fragment thereof.~~

10. ~~The method according to Claim 1, wherein at least one copy of said second DNA sequence is integrated into the genome of said expression host.~~

11. The method according to Claim 10, wherein said expression host further includes at least one copy of a plasmid comprising said second DNA sequence.

12. A method of obtaining an alkalophilic *Bacillus* strain having a reduced extracellular alkaline protease level comprising:

transforming an alkalophilic *Bacillus* strain with a cloning vector comprising a gene coding for a high alkaline protease from which the coding region and optionally portions of the 5' and the 3' non-coding regions have been deleted leaving a sequence of said gene capable of homologous recombination with an indigenous gene coding for a high alkaline protease whereby transformants are obtained;

growing said transformants under conditions whereby the replication function encoded by said vector is inactivated; and

isolating transformants identified as having a reduced extracellular alkaline protease level.

13. The method according to Claim 12, wherein said alkalophilic *Bacillus* strain is *Bacillus* novo species PB92 or a derivative thereof.

14. An alkalophilic *Bacillus* strain capable of producing a mutant high alkaline protease substantially free of expression product of an indigenous protease gene, said strain having been obtained according to the method of transforming a mutant alkalophilic *Bacillus* strain characterized as incapable of producing said expression product with a plasmid expression vector comprising a mutant high alkaline protease gene.

15. The *Bacillus* strain according to Claim 14, wherein said mutant alkalophilic *Bacillus* strain is a mutation of *Bacillus* novo species PB92 or a derivative thereof.

16. The *Bacillus* strain according to Claim 15, wherein said indigenous gene has been deleted by homologous or illegitimate recombination.

Sub B

Sub B

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17. A high alkaline protease substantially free from contamination with a wild-type high alkaline protease, differing in at least one amino acid from the wild-type protease produced according to the method of Claim 1.

not different from any other (substantive)

18. The high alkaline protease according to Claim 17, wherein ~~said Bacillus strain~~ is an alkalophilic Bacillus strain.

no Bac. strain in clm 17  
112

19. A detergent composition containing an active ingredient comprising one or more high alkaline proteases according to Claim 16.

20. Use of one or more high alkaline proteases according to Claims 16 in a detergent composition.

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21. Use of one or more high alkaline proteases according to Claim 16 in a laundry process.

add a'

1/22nd

this is a species used, not a protease.

Add B9

add

Add 1/4